Amendments to the Specification

Replace the paragraph at page 7, lines 2 through 6 with the below paragraph.

Figure 1A-[[1C]] 1D illustrates the nucleotide sequence determined from a genomic clone encoding a human CKR-3 protein also referred to as Eos L2 receptor (SEQ ID NO:1), and the predicted amino acid sequence of the protein encoded by the open-reading frame (SEQ ID NO:2).

Replace the paragraph at page 7, lines 7 through 10 with the below paragraph.

Figure 2A-[[2B]] 2C illustrates the nucleotide sequence determined from the cDNAs encoding a human CKR-3 receptor (SEQ ID NO:3), and the predicted amino acid sequence of the protein encoded by the open-reading frame (SEQ ID NO:4).

Replace the paragraph at page 7, line 32 through page 8, line 3 with the below paragraph.

~~Figure 5~~ Figures 5A-5I [[is]] are an illustration of a FACS analysis of various clones of L1-2 pre-B cells transfected with Eos L2. Cells from over 200 clones were stained with M2 anti-FLAG Mab followed by anti-mouse Ig-FITC. (Y-axis, number of cells; X-axis, fluorescence). In the negative control (PAUL 001), transfected cells were stained with an irrelevant antibody.

Replace the paragraph at page 8, lines 22 through 29 with the below paragraph.

Figures 9A-9D are graphs illustrating CKR-3 expression on leukocytes as determined using MAb LS26-5H12 and flow cytometry. Leukocyte subsets were stained with anti-CKR-3 MAb LS26-5H12 (solid lines) or an IgG_1 isotype-matched control antibody (MOPC-21) (~~shaded profile~~ dotted lines). Figure 9A, eosinophils; Figure 9B, T Cells; Figure 9C, monocytes; Figure 9D, neutrophils. Dead cells were excluded based on propidium iodide staining.

Replace the paragraph at page 8, line 30 through page 9, line 3 with the below paragraph.

Figures 10A-10C are graphs illustrating cell surface staining of L1.2 cells transiently transfected with a CKR-3 receptor (Figure 10A), mock-transfected L1.2 control cells (Figure 10B), or cell line E5 (a stable L1.2 CKR-3 transfectant) (Figure 10C) with an anti-CKR-3 monoclonal antibody (LS26-5H12, solid line). Background staining with control monoclonal antibody MOPC-21 is also shown (~~shaded profile~~ dotted lines).

Replace the paragraph at page 9, lines 4 through 14 with the below paragraph.

Figures 11A-11D are graphs illustrating the results of competitive ligand binding of radiolabeled human eotaxin to the E5 cell line (a stable L1-2 cell line transfected with a CKR-3 receptor; Figure 11A) or to human eosinophils (Figure 11B). Cells were incubated with 0.6 nM ^{125}I-labeled eotaxin and various concentrations of unlabeled eotaxin (O), RANTES ([[Δ]] ▲), or MCP-3 (□). After 60 minutes at room temperature, cell pellets were washed and counted. Scatchard plots of unlabeled eotaxin competition were calculated from the data (Figure 11C, E5 cell line; Figure 11D, eosinophils).

Replace the paragraph at page 10, lines 9 through 14 with the below paragraph.

Figure 15 is a graph illustrating the binding of ^{125}I-labeled RANTES to [[a]] membranes from a stable cell line (A31-293-20) obtained by transfecting 293 cells with the A31 cDNA clone (square with central dot) as compared with binding to membranes from untransfected 293 cells (filled circles).

Replace the paragraph at page 11, lines 13 through 23 with the below paragraph.

Figure 19 is a graph illustrating inhibition of binding of radiolabeled eotaxin, RANTES, and MCP-3 to human eosinophils by mAb 7B11. Human eosinophils were incubated with

0.1 nM ^{125}I-labeled-eotaxin, -RANTES, or -MCP-3, and various concentrations of mAb 7B11. After 60 minutes at room temperature, cell pellets were washed and counted. Data was analyzed by ~~KaleidaGraph~~ KALEIDAGRAPH software, which calculated an IC50 [[of]] for eotaxin of 25.7 ng/ml, for RANTES of 13.7 ng/ml, and for MCP-3 of 18.8 ng/ml. The level of inhibition using 250 nM cold chemokine is shown at the bottom left of the plot: [[●]] ○ eotaxin, [[□]] □ RANTES, and [[▲]] △ MCP-3.

Replace the paragraph at page 12, lines 4 through 18 with the below paragraph.

~~Figure 21~~ Figures 21A-21J [[is]] are a series of tracings illustrating that mAb 7B11 inhibits $[Ca^{2+}]i$ by human eosinophils in response to eotaxin, RANTES, MCP-2, MCP-3 and MCP-4. Human eosinophils were labeled with Fura-2, and stimulated sequentially with mAb (A), followed 40 sec later with the indicated chemokine (B), and 100 sec following that with C5a (C). $[Ca^{2+}]i$ fluorescence changes were recorded using a spectrofluorimeter. The tracings are representative of five separate experiments, performed with eosinophils from different donors. In the top panels, an irrelevant control mAb (MOPC-21) was used, and in the bottom panels, mAb 7B11. Antibodies were used at a final concentration of 6.4 μg/ml Chemokines were used at: eotaxin, 10 nM, RANTES, 20 nM, MCP-2, 200 nM, MCP-3, 200 nM, MCP-4, 10 nM. C5a was used at 400 pM.

Replace the paragraph at page 13, lines 7 through 15 with the below paragraph.

Figure 23A is a histogram illustrating blockade of eotaxin-, RANTES- and MCP-3-induced eosinophil peroxidase (EPO) release by monoclonal antibody 7B11. ~~Clear~~ Cross hatched bars indicate the amount of EPO released by either 10 nM eotaxin, 100 nM eotaxin, 100nM RANTES or 100 nM MCP-3. Black bars indicate the amount of EPO released when 10 μg/ml of 7B11 was present in the eosinophil degranulation assay. The bar marked "blank" corresponds to a no chemokine, no antibody (buffer) control.

Replace the paragraph at page 13, lines 16 through 21 with the below paragraph.

Figure 23B is a histogram illustrating the effect of mAb 7B11 on C5a-induced eosinophil peroxidase release. ~~Clear bars indicate~~ The cross hatched bar indicates the amount of EPO released by 1 nM C5a. The black bar indicates ~~Black bars indicate~~ the amount of EPO released when 10 μg/ml of 7B11 was present in the eosinophil degranulation assay.

Replace the paragraph at page 14, lines 10 through 12 with the below paragraph.

Figure 30B is a histogram illustrating blockade of basophil ~~chemotax is~~ chemotaxis in response to eotaxin and MCP-4 using anti-CCR3 mAb 7B11.

Replace the paragraph at page 14, lines 14 through 27 with the below paragraph.

As described herein, nucleic acids encoding a novel human receptor, designated Eos L2 or C-C chemokine receptor 3 (CKR-3), also referred to herein as "CCR3", have been isolated. Both human genomic and cDNA clones have been characterized. The cDNA clone was isolated from an eosinophil cDNA library constructed from eosinophils obtained from a patient with hypereosinophilic syndrome. Sequence analysis of the clones revealed a gene containing an open reading frame of 1065 nucleotides encoding a predicted protein of 355 amino acids (Figures 1A-[[1C]] 1D and 2A-[[2B]] 2C; SEQ ID NOS: 2 and 4), which shares amino acid sequence similarity with other C-C chemokine receptors, which are believed to be G protein-coupled receptors and to have a similar structure of seven transmembrane spanning regions.

Replace the paragraph at page 44, lines 21 through 35 with the below paragraph.

An isolated, recombinant mammalian CKR-3 receptor protein, such as a human CKR-3 receptor as that shown in Figure 1A-[[1C]] 1D (see also, SEQ ID NO:2), Figure 2A-[[2B]] 2C (see also, SEQ ID NO:4) or SEQ ID NO:6, can be used in the present method, in which the effect of a compound is assessed by monitoring receptor function as described herein or using other

suitable techniques. For example, stable or transient transfectants, such as A31/293/#20 stable transfectants (see e.g., Example 9), stable tranfectants of mouse L1-2 pre-B cells (see e.g., Example 3), baculovirus infected Sf9 cells (see e.g., Example 4), can be used in binding assays. Stable transfectants of mouse L1-2 pre-B cells or of other suitable cells capable of chemotaxis can be used (see e.g., Example 3) in chemotaxis assays, for example.

Replace the paragraph at page 54, lines 12 through 17 with the below paragraph.

Chambers can be formed from various solids, such as plastic, glass, polypropylene, polystyrene, etc. Membranes which are detachable from the chambers, such as a ~~Biocoat~~ BIOCOAT (Collaborative Biomedical Products) or ~~Transwell~~ TRANSWELL (Costar, Cambridge, MA) culture insert, facilitate counting adherent cells.

Replace the paragraph at page 72, lines 2 through 17 with the below paragraph.

Chemokines were obtained from Peprotech, Inc. (Rocky Hill, N.J.). Chemotaxis experiments were performed using 3.0 micron ~~Biocoat~~ BIOCOAT cell culture inserts (Collaborative Biomedical Products), in 24 well plates. Endothelial cells were grown to confluency on the inserts for two days prior to chemotaxis experiments. The endothelial cells used were a cell line termed ECV 304 (European Collection of Animal Cell Cultures, Porton Down, Salisbury, U.K.), which expresses endothelial cell markers such as von Willebrand factor, as well as ICAM-1 and VCAM-1. This endothelial cell line greatly facilitates these assays, since human umbilical vein endothelial cells can be variable in nature, can be used for only several passages, and grow much more slowly than ECV 304. The assay was conducted at 37°C for 1.5 hours, and migrated cells were counted using an inverted microscope.

Replace the paragraph at page 91, line 32 through page 92, line 6 with the below paragraph.

MAbs reactive with the Eos L2 receptor were generated by immunizing mice with a synthetic peptide corresponding to the N-terminal 35 amino acids. The N-terminal 35 amino acids of Eos L2, deduced from the nucleotide sequence (see Figures 1A-[[1C]] 1D; see also, SEQ ID NO:2), were synthesized and coupled to the carrier protein PPD (Purified Protein Derivative of *Mycobacterium tuberculosis*; Severn Biotech Ltd., Cambridge, U.K.).

Replace the paragraph at page 95, lines 3 through 16 with the below paragraph.

2% - 5% of transiently transfected COS, HEK-293 and CHO cells were surface positive as assessed using antibodies to FLAG-tagged receptor (see above), while substantial intracellular protein could be detected, suggesting inefficient protein trafficking. The L1.2 mouse pre-B cell line was used to select lines with higher levels of surface expression (see ~~Figure 5~~ Figures 5A-5I) for further assessment of ligand binding specificity and signal transduction by CKR-3. This cell line has been used successfully for the study of other chemoattractant receptors (Honda, S., *et al., J. Immunol., 152*: 4026-4035 (1994)), and the expression of transfected human chemokine receptors confers specific chemotactic ability to various ligands (see below).

Replace the paragraph at page 100, lines 3 through 16 with the below paragraph.

Competition is presented as the percentage of specific binding as calculated by 100(S-B)/(T-B), where S is the radioactivity of the sample, B as background binding and T as total binding without competitors. Background binding was obtained by incubating cells with radiolabeled chemokine and at least 400-fold excess of unlabeled chemokines. The total binding of eotaxin to E5 cells was 11611 ± 119 cpm and background binding 2248 ± 745 cpm. The total binding of eotaxin to eosinophils was 7866 ± 353 cpm and background binding 1148 ± 518 cpm. Duplicates were used throughout the experiments and the standard deviations were always less

than 10% of the mean. All experiments were repeated at least three times. Curve fit was calculated by ~~KaleidaGraph~~ KALEIDAGRAPH software.

Replace the paragraph at page 106, lines 10 through 24 with the below paragraph.

The nucleotide sequence of the full-length cDNA and the predicted amino acid sequence of the encoded protein are shown in Figures 2A-[[2B]] 2C (see also SEQ ID NO:3 and SEQ ID NO:4). The cDNA sequence shown in Figures 2A-[[2B]] 2C was determined from clones A31 (bases 15-365 (numbering as in Figures 2A-[[2B]] 2C)), and the M-16/Bluescript construct (bases 366 to 1152 (numbering as in Figures 2A-[[2B]] 2C)). A comparison of the amino acid sequence of the novel receptor with other proteins revealed that the novel receptor and the MIP-1α/RANTES receptor share 62% sequence identity, and the novel receptor and the MCP-1 receptor share 50.57% sequence identity. Sequence identity was determined using the Wisconsin UW GCG package (program gap), with the Needleman and Wunsch algorithm (Needleman and Wunsch, *J. MOl. Biol. 48*:443-453 (1970)).

Replace the paragraph at page 116, lines 12 through 22 with the below paragraph.

^{125}I-labeled eotaxin was obtained from Amersham (Arlington Heights, IL), and its specific activity was stated to be 2000 Ci/mM. Chemokine binding to target cells was carried out as described previously (Ponath, P.D., *et al., J. Clin. Invest., 97*:604-612 (1996); Van Riper, G., *et al., J. Exp. Med., 177*: 851-856 (1993)). Duplicates were used throughout the experiments and the standard deviations were always <10% of the mean. All experiments were repeated at least three times. Curve fit and concentrations that inhibit 50% specific binding (IC50) were calculated by ~~KaleidaGraph~~ KALEIDAGRAPH software (Synergy Software, Reading, PA).

Replace the paragraph at page 119, lines 15 through 27 with the below paragraph.

MAb 7B11 inhibits changes in [Ca^{2+}]i by eosinophils in response to cc chemokines. Eotaxin, RANTES, MCP-2, MCP-3 and MCP-4 induce changes in [Ca^{2+}]i by human eosinophils

(Ponath, P.D., *et al., J. Clin. Invest., 97*:604-612 (1996); [[(]] Uguccioni, M., *et al., J. Exp. Med., 183*:2379-2384 (1996)). To examine the agonist/antagonist function of mAb 7B11, eosinophils were assessed for $[Ca^{2+}]i$ following injection of mAb 7B11, or an irrelevant control mAb. Eosinophils incubated with the irrelevant mAb still produced changes in $[Ca^{2+}]i$ following injection of optimal amounts of eotaxin, RANTES, MCP-2, MCP-3 and MCP-4 (~~top panels, Figure 21~~ Figures 21A, 21C, 21E, 21G and 21I). C5a, a potent stimulator of eosinophil $[Ca^{2+}]i$, was used as a control.

Replace the paragraph at page 119, line 28 through page 120, line 7 with the below paragraph.

Eosinophils incubated with 6.4 μg/ml of 7B11 mAb for 40 seconds were unable to respond to eotaxin, RANTES, MCP-2, MCP-3 and MCP-4 (~~bottom panels, Figure 21~~ Figures 21B, 21D, 21F, 21H and 21J). This inhibition was not due to receptor modulation from the cell surface, since this effect was rapid, and immunofluorescent staining of eosinophils incubated with mAb 7B11 at room temperature revealed intense staining. In addition, mAb7B11 was antagonistic rather than agonistic, since concentrations as high as 10 μg/ml of mAb failed to induce a change in $[Ca^{2+}]i$. 7B11 treated eosinophils showed changes in $[Ca^{2+}]i$ to C5a ~~(Figure 21)~~. mAb 7B11 had no effect on the $[Ca^{2+}]i$ of butyrate differentiated HL-60 cells to MIP-1α or RANTES, a response that is mediated through receptors other than CCR3.

Replace the paragraph at page 120, lines 8 through 32 with the below paragraph.

IL-5 primed eosinophils respond to CC chemokines through CCR3 but upregulate IL-8 receptors. Eosinophils from eosinophilic individuals, and normal eosinophils primed *in vitro* with IL-5, respond to IL-8 in chemotaxis assays (Schweizer, R.C., *et al., Blood, 83*:3697-3704 (1994); Sehmi, R., *et al., Clin. Exp. Allergy, 23*: 1027-1034 (1994)), suggesting that activated eosinophils have altered chemokine receptor expression. To test whether primed or activated eosinophils respond to CC chemokines in the same manner as do normal eosinophils,

blocking experiments similar to those shown in Figures 20 and 21A-21J were performed using day 5 to 7 IL-5 stimulated eosinophils, and eosinophils from an eosinophilic individual. The IL-8 receptors, CXCR1 and CXCR2, were undetectable by mAb staining on eosinophils from all normal individuals examined (n=12) (Figure 22A). However following 5-7 days in culture in vitro with human IL-5, CXCR2 and (to a lesser degree) CXCR1 were detectable on the surface of eosinophils, as detected using anti-CXCR2 mAbs and flow cytometry (Figure 22B), and this expression paralleled the ability of these eosinophils to migrate to IL-8 in chemotaxis assays (not shown). In the one eosinophilic donor examined (18-25% of WBC were eosinophils, for >1 year), CXCR2 was expressed on eosinophils at a slightly lower level (Figure 22C).